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Species

Myxobolus deformis sp. nov. (myxozoa, myxosporea, myxobolidae), a new myxosporean parasite infesting the gills of *Cyprinus carpio*.

Naireen Fariya[1]☼, Rehana Abidi[2], Chauhan UK[3]

1.Research Scholar, Parasitology Lab, Fish Health Management Division, National Bureau of Fish Genetic Resources, Canal Ring Road, Dilkusha, Telibagh, Lucknow 226002, India
2.Principal Scientist, Parasitology Lab, Fish Health Management Division, National Bureau of Fish Genetic Resources, Canal Ring Road, Dilkusha, Telibagh, Lucknow 226002, India
3.School of Environmental Sciences, A.P.S. University, Rewa, M.P., India

☼**Corresponding Author:**
Dr. Naireen Fariya,
Ground Floor, Hasan Vila, H. NO. Q-7 (Old No. F-56/29),
Batla House, Near Hari Masjid, Jamia Nagar,
Okhla, New Delhi – 110025,
India.
Email: n_fariya@yahoo.com

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ABSTRACT

Cyprinus carpio is a commercially important food fish widely cultured in India. It has high nutritive value as a food and has great economic importance in fish trade. The economic value of fish as a food is reduced by infectious diseases. Among susceptible diseases particularly parasitic infestations are distressing. *Cyprinus carpio* is vulnerable to various parasitic diseases but myxozoan infestations are one of the common problem. Present study reveals a new species of *Myxobolus* in *Cyprinus carpio* named as *Myxobolus deformis* sp. nov infecting the gills. There were no cyst or plasmodia but minute spores were embedded in the gill filaments. The spores were almost spherical but spore length is shorter than the width.This article deals with morphological and morphometric description of *M. deformis* sp. nov. Morphological details of the mature spores were compared with the similar spores as well as species previously reported from *Cyprinus carpio*.

Key Words - *Cyprinus carpio*, Exotic carp, *Myxobolus deformis*, Myxosporean, Parasite.

Abbreviations: LS- Length of Spore; WS- Width of Spore; LPC- Length of Polar Capsule; WPC- Width of Polar capsule; SD- Standard Deviation.

1. INTRODUCTION

Cyprinus carpio is an exotic carp generally known as common carp in local market. It is one of the thriving species for cultivation in this region (Srivastava & Singhal, 2015). *Cyprinus carpio* is widely cultured as food and ornamental fish. The economic value of fish as a food is reduced by infectious diseases. Lafferty et al., (2015) stated that most infectious diseases in farmed fishes are introduced by wild species and same can be transferred to the wild species. Various parasites cause severe diseases to wide range of fishes including both cultured and wild. Parasitic infestation particularly Myxozoan causes serious injuries and high mortalities to freshwater fishes (Abidi et al. 2015). Among myxozoans, the genus *Myxobolus* includes the highest number of species. Genus *Myxobolus* are very important and fatal parasites because they infect economically important fish species and cause high mortality in farmed fish (Fiest, 2008). Several infections caused by *Myxobolus* species in *C. carpio* are reported by various workers worldwide (Eiras et al. 2005; 2014).

In India, more than 104 species of *Myxobolus* have been recorded infecting freshwater and marine fishes (Kalavati & Nandi 2007). Further, a revised synopsis of 131 nominal species of *Myxobolus* from India was reported by Kaur & Singh (2012). While Eiras et al., in 2014, listed 112 species of *Myxobolus*, out of which 29 species were from India. New myxosporean parasites are constantly emerging and are causing serious threat to the development of the pisciculture (Kaur 2014). Research on myxosporean fish parasites is a fast developing field of Ichthyo-parasitology. With this view, the present study was finished and the currentarticle gives the description of a new pathogenic myxosporean parasite infesting the gills of Chinese carp, *Cyprinus carpio*.

2. MATERIAL AND METHODS

Fishes were collected from river Gomti and Kaisarbagh fish market of Lucknow. Thorough investigation of body surface, fins, gills and internal organs were done for the presence of Myxospores. Gills were infected with numerous spores of Myxobolus. No cysts were found. Fresh spores were examined under a Nikon E600 microscope with various magnifications (including oil immersion) and treated with 12% KOH solution for the extrusion of polar filaments. For permanent preparations, air-dried smears were stained with Geimsa and Leishman's stains (Fig2.a, b). Drawings were made from fresh and stained material with the aid of Camera Lucida and Corel Draw 17.0 software (Fig3.a, b, c). Morphometric measurements based on65 fresh spores were done with the help of software NIS-E- Br. All measurements are taken in micrometers with ranges values followed by mean ± SD in parentheses.

3. RESULTS

Parasite: *Myxobolus deformis* sp. nov

Parasite Profile

Host:	*Cyprinus carpio*
Locality:	Lucknow, Uttar Pradesh, India**.**
Site of infection:	Gills

Prevalence of infection: 9.61%

Etymology:

The species is named *M. Deformis* because it is different from usual shape of other *Myxobolus* species morphologically and seems to be pressed &deformed shaped comparatively.

Prevalence:

Total 52 *Cyprinus carpio* were examined thoroughly, out of which 5 fishes were found infected with *Myxobolus deformis* sp. nov. Several histozoic, minute spores were observed in the gills. The intensity of infection was high, but the prevalence of the infection was 9.61%. There was no cyst or plasmodia in the gills.

Description:

The spores are almost spherical but have an unusual shape as spore width was exceeded in comparison to the length (i.e. spore length was shorter than the width) comprising mean length was 7.01 ± 0.48 µm (range 5.81-7.94 µm) and mean width was 9.39±0.57 µm (range 8.09- 10.65 µm) (Fig. 4-6). In frontal view, spores were short, blunt, stout, more or less spherical, dorso-ventrally flattened (Fig. 1a). Spore had a thick cell wall. The polar capsules were also thick walled, drop-shaped and occupy most of the spore's space. The size of polar capsules consists, mean length 4.53 ± 0.57 µm (ranging 3.25- 5.80µm) and mean width 3.31± 0.40µm (ranging 2.12- 4.15µm) (Fig. 7, 8) (Table - I). There was a slight difference in the size of two capsules, which was also recorded separately as larger capsule's mean length was 4.76 ± 0.53µm (ranging 3.85-5.8 µm) and mean width was 3.55 ± 0.32µm (ranging 2.72-4.15 µm). As well, smaller capsule's mean length was 4.30 ± 0.52µm (ranging 3.25-5.44 µm) and mean width was 3.07 ± 0.33µm (ranging 2.12-3.79µm). Polar were also of unequal length after full extrusionand coils 3 - 5 times (Fig1. b). The mean length of extruded bigger filament is 45.19 ± 18.62µm (range- 27.68 - 76.14µm) and smaller filament is 32.50 ± 13.34µm (range - 20.71-58.72µm). In sutural view, spores are nearly oval or lenticular and covered by shell valves. Sporoplasm is homogenous and entirely filled in the extra-capsular space below the capsules (Fig1.a).

Table I

Morphometric characteristics of *Myxobolus deformis* sp. nov

Characters	Range in µm	Mean in µm	St. deviation in µm
LS	5.81-7.94	7.01	0.48
WS	8.09- 10.65	9.39	0.57
LPC	3.25- 5.80	4.53	0.57
WPC	2.12- 4.15	3.31	0.40
Ratio: LS/WS	-	0.75	-

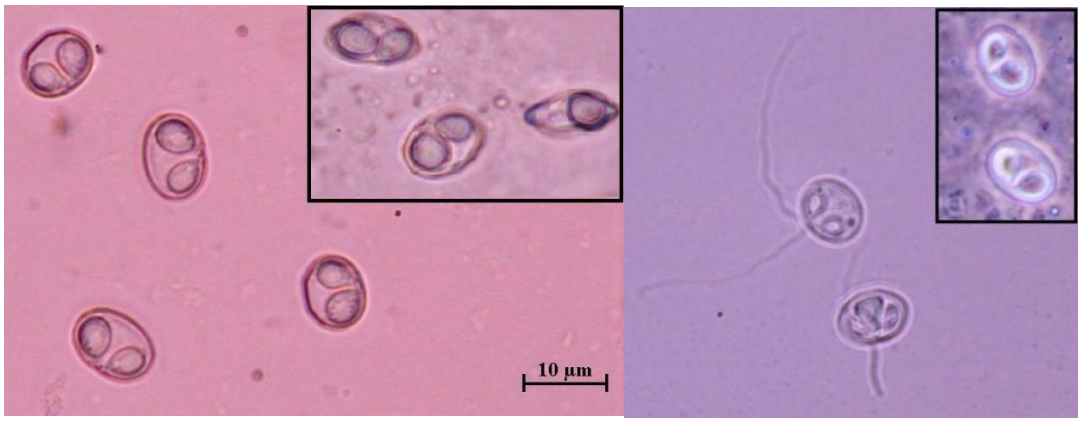

Figure 1

(a) Mature Spores of *Myxobolus deformis* sp. nov.(b) Mature Spores of *Myxobolus deformis* sp. nov with extruded filament

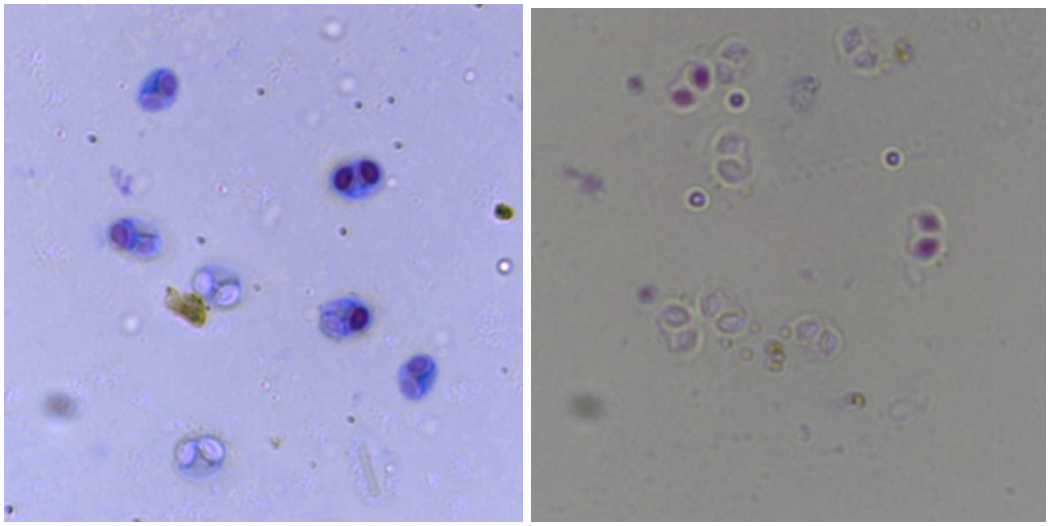

Figure 2

(a) Myxopores of *Myxobolus deformis* sp. nov stained with Geimsa. (b) Myxopores of *Myxobolus deformis* sp. nov stained with Leishman's stain

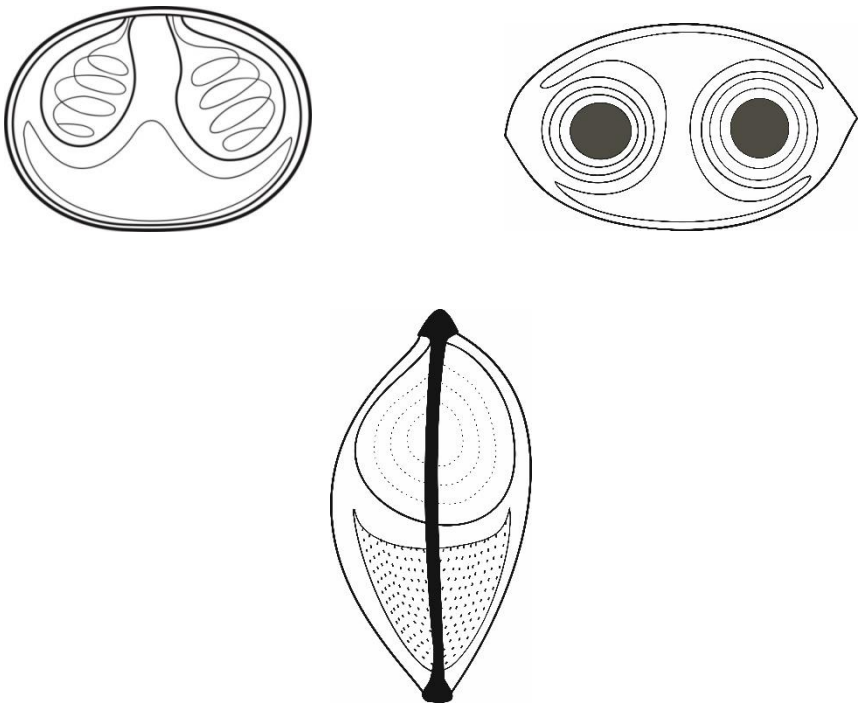

Figure 3

(a) Line drawing of fresh mature myxospore of *Myxobolus deformis* sp. nov.infecting gills of *C. carpio* (Frontal view) (b) Line drawing of fresh mature myxospore of *Myxobolus deformis* sp. nov.infecting gills of *C. carpio* (Lateral view) (c) Line drawing of fresh mature myxospore of *Myxobolus deformis* sp. nov. infecting gills of *C. carpio* (Sutural view)

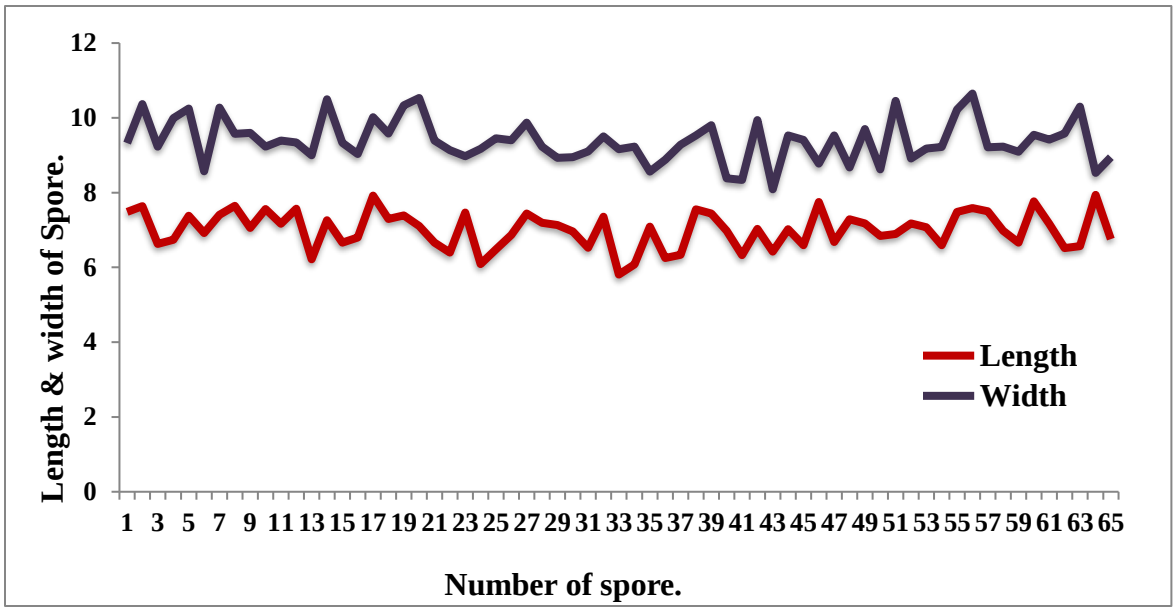

Figure 4
Graph displaying Spore Length & Width of *M. deformis* sp. nov.

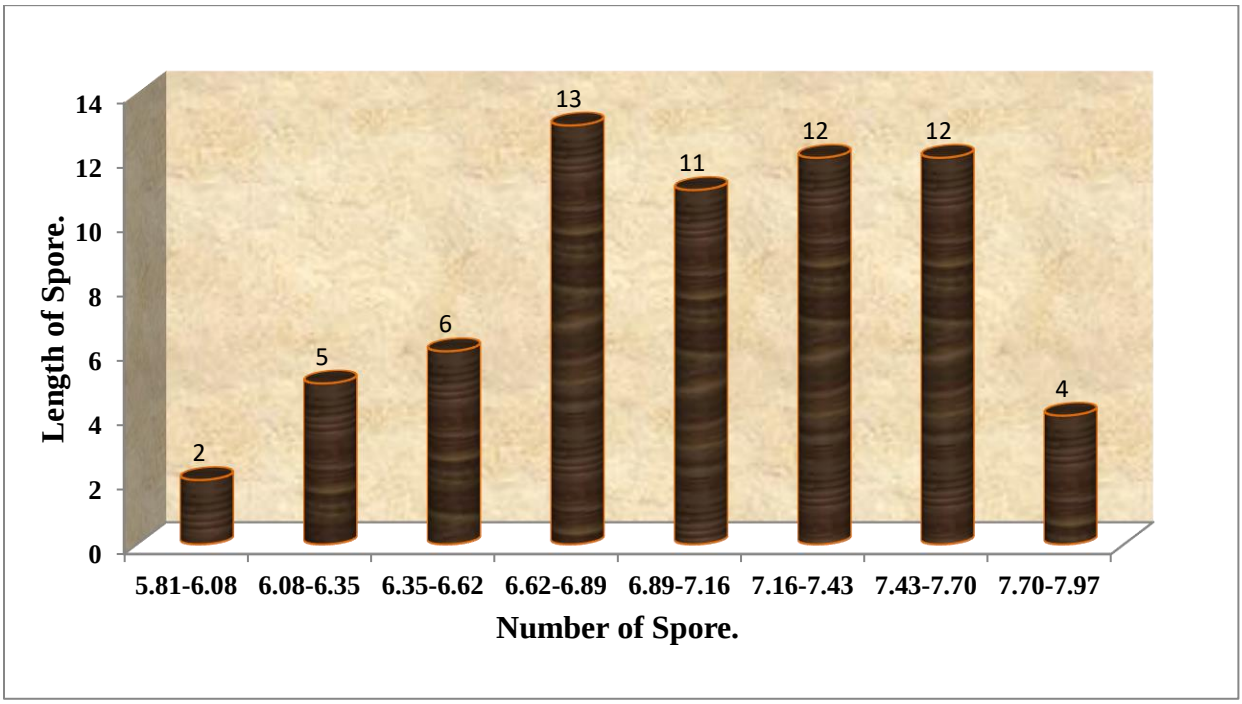

Figure 5
Histogram showing Spore Length of *M. deformis* sp. nov.

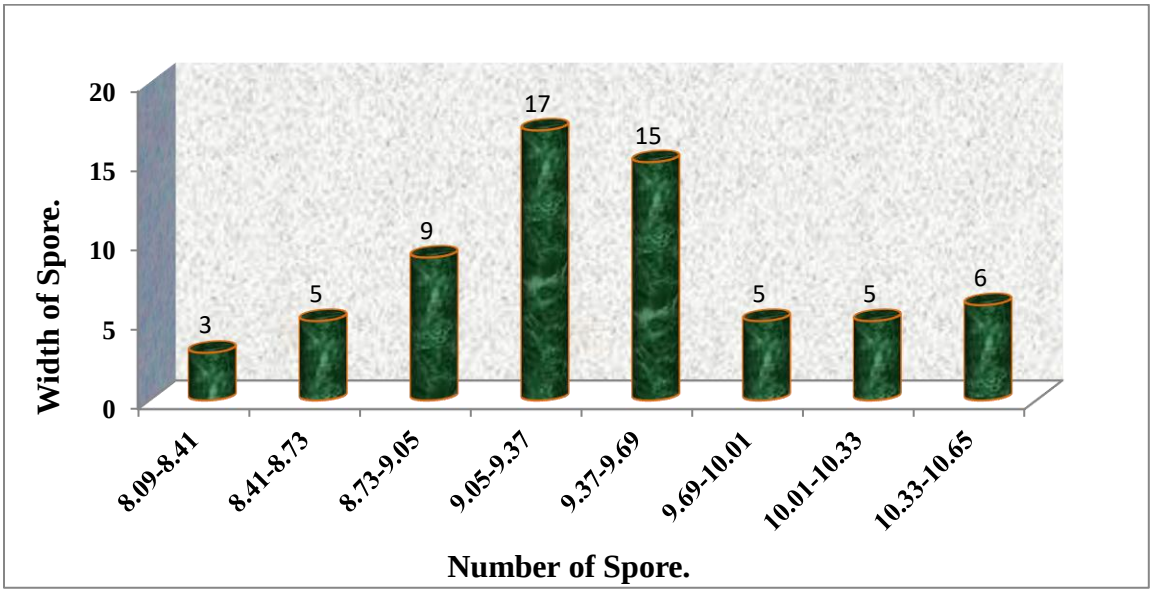

Figure 6
Histogram showing Spore Width of *M. deformis* sp. nov.

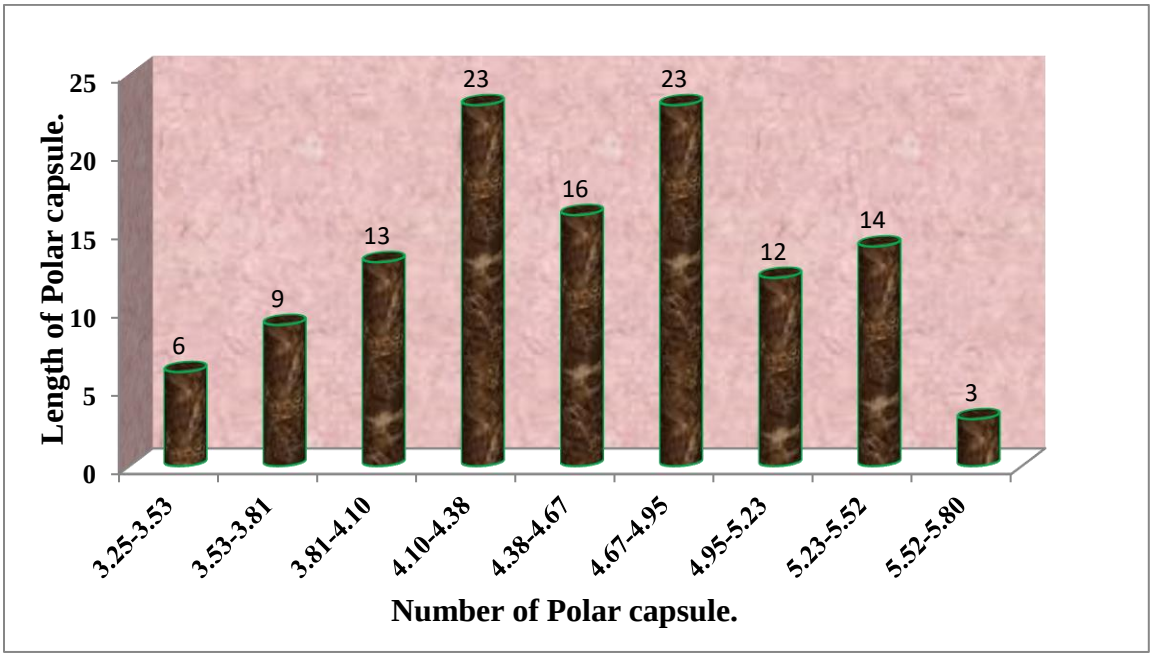

Figure 7
Histogram showing Polar capsule Length of *M. deformis* sp. nov.

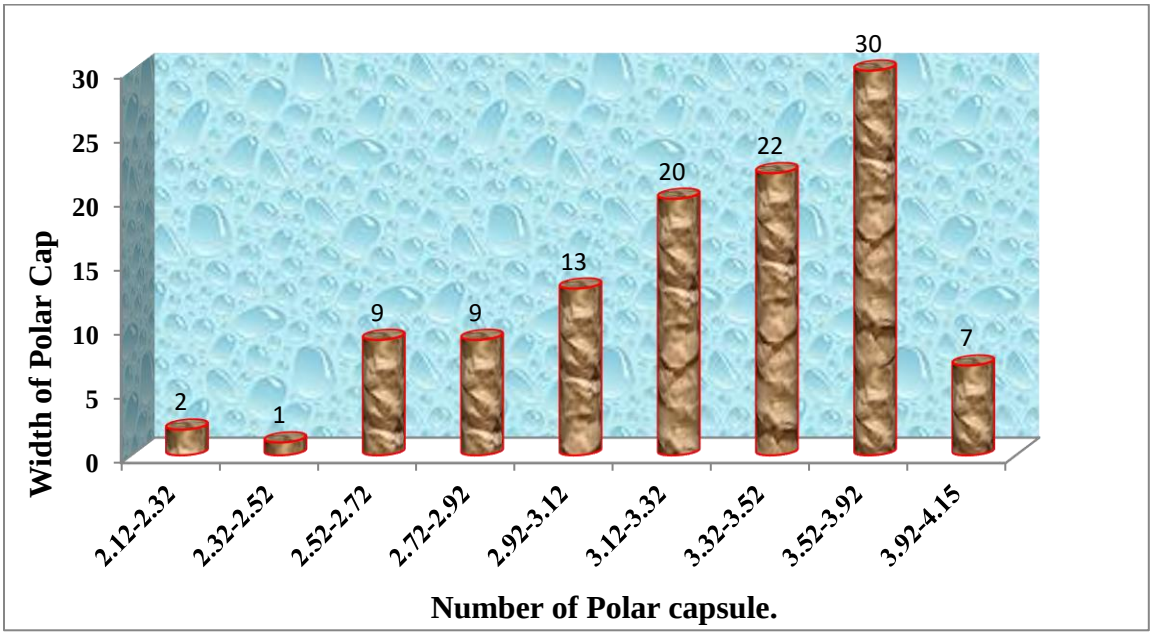

Figure 8
Histogram showing Polar capsule Width of *M. deformis* sp. nov.

4. DISCUSSION

The present new species *M. deformis* sp. nov was compared with all the previously known species from isolated from *Cyprinus carpio* by earlier workers. These species include *M. amurensis* from fins, gut (Akhmerov, 1960); *M. basilamellaris* from gills (Lom & Molnar, 1983); *M. cuttacki* from gills (Haldar et al. 1996); *M. cyprini* from muscles (Doflein, 1898); *M. dispar* from gills (Thelohan, 1895); *M. encephalicus* from brain (Mulsow, 1911; Landsberg & Lom, 1991); *M. hanchuanensis* from gills, body-cavity (Chen & Ma, 1998); *M. heteromorpha* from heart, kidneys (Ma {a, b} 1993); *M. intrachondrealis* from cartilage of gill arches (Molnar, 2000); *M. junchisi* from gills, muscles, kidneys (Yukhimenko, 1986); *M. longisporus* and *M. microlatus* from almost all organs (Nie & Li, 1973; Nie & Li, 1992); *M. miyunensis* from kidneys (Chen & Ma, 1998); *M. musseliusae* from gills (Yakovchuk 1979); *M. acinosus* from gills, *M. nielii* from almost all organs (Nie & Li, 1973; Landsberg & Lom, 1991); *M. paratoyamai* from nares, ureter (Nie & Li, 1992); *M. rotundatus* from gut (Akhmerov, 1956); *M. sinocyclochilusi* from gills (Ma, 1998); *M. wuchangensis* from caudal fins (Chen & Ma, 1998); *M. wucheni* from kidneys, gills (Wu & Chen, 1987; Landsberg & Lom, 1991); *M. yibinensis* from muscle (Zhao & Ma, 1994); *M. cyprinicola* from intestine (Reuss, 1906); *M. elliptoides* from fins (Wu & Chen, 1987); and *M. serratus* from gill arch (Pagarkar & Das, 1993 {emend}). All above species are different morphologically and morphometrically from present species.

Further the present species were compared with *Myxobolus* sp. from other hosts such as *M. Lalbaghensis* from gills of *Labeobata* (Banerjee et al. 2011); *M. Analfinus* from anal fin of *Heteropneustes fossilis*; *M. debsantus* from tail fin of Hybrid carp of *Catla-Rohu* (Basu et al. 2009); *M.calcariferum* from gill lamellae of *Latescalcarifer*; *M. chinsurahensis* from scales of *Anabas testudineus*; *M. Mrigalhitae* from gill filaments of Hybrid carp of *C. mrigala - L. rohita* (Basu & Haldar 2003); *Myxobolus sp.n* PKB2014 from gill lamellae of *Labeorohita* (Panda et al. 2015). But all the above species were dissimilar in spore and polar capsule, to the present spores which possesses a unique in shape with broader in width and short in length.

Table II
Comparative description of *Myxobolus deformis* sp. nov.

Species	Host	Site of Infection	Spore		Polar Capsule		References
			Length(µm)	Width (µm)	Length(µm)	Width (µm)	
Myxobolus deformis sp. nov	*Cyprinus carpio*	Gills	7.01 (5.81-7.94)	9.39 (8.09-10.65)	4.76 (3.85-5.80); 4.30 (3.25- 5.44)	3.55 (2.72-4.15); 3.07 (2.12- 3.79)	Present paper
M. amurensis	*Cyprinus carpio*	fins, gut	9–13.5	9–12.5	4.5–7	3.8–4.2	

M. basilamellaris	*Cyprinus carpio*	gills	7.7–12.2	7.3–9.9	3.2–5.4	2.2–3.3	
M. cuttacki	*Cyprinus carpio Cyprinus carpio*	gills	17.0 (13.0-21.1)	6.4 (4.9–8.1)	8.6 (6.5–13)	2.8 (1.6–4.0)	
M. cyprini		muscles	10–16	8–12	5.2–7	--	
M. dispar	*Cyprinus carpio*	gills	10–12	8	7	5	
M. encephalicus	*Cyprinus carpio*	brain	5–5.5	5–5.5	--	--	
M. hanchuanensis	*Cyprinus carpio, Rhodeussinensis*	gills, body-cavity	13.0 (12.0-13.7)	10 (9–11)	4.5 (4–4.8)	2.8 (2.6–3)	Eiras et al. 2005
M. heteromorpha	*Cyprinus carpio*	heart, kidneys	10.9 (9.1–11.8)	9.5 (8.8–10.3)	5.2 (4.4–5.9)	3.6 (2.9–4.4)	
M. intrachondrealis	*Cyprinus carpio*	cartilage of gill arches	10.2 (9.0–11)	6.5 (6–7)	4.5 (3.7–4.7)	2.2 (2–2.6)	
M. junchisi	*Cyprinus carpio haematopterus*	gills, muscles, kidneys	9.7–12.6	8.4–9.2	5.4–6.3	2.9–3.2	
M. longisporus	*Cyprinus carpio*	--	16.0–17.5	6.5 –7 .0	7.5–8.2	2.0	
M. microlatus	*Cyprinus carpio*	almost all organs	8.2 (7.2–8.4)	10 (9.6–11.4)	5.0 (4.2–6.0)	3.8 (3.6–4.0)	
M. miyunensis	*Cyprinus carpio*	kidneys	11.7 (10.8–12)	9.2 (8.4–9.6)	5.9 (5.4–7.2)	2.6 (2.4–3.0)	
M. musseliusae	*Cyprinus carpio*	gills	10.5–11.1	8.8–10	3.9–4	--	
M. acinosus	*Cyprinus carpio haematopterus*	gills	12.6 (10.8–13.2)	6.4 (5.6–7.2)	5.3 (4.8–6.0)	2.8 (2.4–3.4)	
M. nielii	*Cyprinus carpio*	almost all organs	10 (8–12)	8.6 (8.4–9.6)	4.7 (4.2–5.0)	2.9 (2.4–3.0)	
M. paratoyamai	*Cyprinus carpio*	nares, ureter	12.5–14.2	5.5–7.0	6.2–7.4	2.2–2.5	
M. rotundatus	*Cyprinus carpio haematopterus*	gut	8–11	8–11	4.5–5	3–4	
M. sinocyclochilusi	*Cyprinus carpio*	gills	12.7 (11–14.4)	9.9 (8.8–11)	4.9 (4.8–5.2)	2.9 (2.8–3.2)	
M. wuchangensis	*Cyprinus carpio*	caudal fins	10.7 (9.6–11.0)	9.1 (8.4–10.2)	4.6 (4.6–4.8)	2.6 (2.4–2.8)	Eiras et al. 2005
M. wucheni	*Cyprinus carpio*	kidneys, gills	13.5 (12.9–14.3)	10.1 (9–11.2)	5.8 (5.2–7.1)	4.3 (2.4–4.8)	
M. yibinensis	*Cyprinus carpio*	muscles	9.0 (8.5–9.8)	10.9 (10–12)	4.8 (4.6–5.0)	3.5 (3.0–4.2)	
M. cyprinicola	*Leuciscuswaleckii, Cyprinus carpio*	intestine	11–12.2 (11.8)	8.1–9.4 (9.0)	4.8–5.2 (5.0)	3.0–3.4 (3.2)	
M. elliptoides	*Cyprinus carpio*	fins	14.0–15.9	8.8–10.7	4.2–5.9; 2.3–3.5	2.8–3.5; 2.2–2.6	Eiras et al. 2014
M. lalbaghensis	*Labeobata*	gills	7.65- 11.9 (9.22 ±1.0)	5.1-8.5 (6.8 ± 1.17)	4.25-6.8 (5.185 ± 0.706)	1.7- 2.55 (2.337 ± 0.368)	Banerjee et al. 2011
M. analfinus	*Heteropneustes fossilis*	Anal fin	12.3(11.1–13.4)	8.6 (7.8–9.3)	4.1 (3.2–4.9) 2.5 (2.0–3.1)	2.2 (2.0-2.4) 1.8 (1.6–2.0)	
M. debsantus	Hybrid carp *Catla-Rohu*	Tail fin	9.0 (8.5–9.6)	8.4 (8.1–8.9)	4.3 (4.0–4.6) 2.8 (2.6–2.9)	2.3 (2.0–2.6) 1.8 (1.6–1.9)	Basu et al. 2009

M. calcariferum	*Latescalcarifer*	gill lamellae	6.6 (6.1-7.1)	6.2 (5.7-6.5)	4.2 (3.8-4.5)	2.3 (2.0-2.7)	
M. chinsurahensis	*Anabas testudineus*	scales	8.4 (8.0-9.7)	5.4 (5.1-6.1),	4.4 (3.9-6.6)	2.1 (1.8-2.5)	Basu & Haldar 2003
M. mrigalhitae	Hybrid carp *C. mrigala - L. rohita*	gill filaments	10.8 (10.2-11.3)	7.9 (7.6-8.1)	4.8 (4.3-5.2)	2.9 (2.7-3.2)	
*Myxobolus sp.n*PKB2014	*Labeorohita.*	gill lamellae	14.7 (11.1-19.1)	6.36 (5.3-7.3)	10.64 (8.2-14.2) 9.55 (7.1-12.9)	2.99 (2.3-3.9) 2.79 (2.2-3.6)	Panda et al. 2015
M. brachysporus	*Tilapia esculenta, T. variabilis*	Spleen	7.3 (7.0–7.5)	12.5 (12–13.5)	3.1 (2.5–3.8)	2.3 (2.3–2.5)	Eiras et al. 2005
M. brachysporus	*Oreochromis niloticus*	Spleen	8.6 (7.8 - 9.2)	13.2 (12.1-14.2)	4.7 (4.2-5.1)	3.6 (3.2 – 4.2)	Abdel-Baki et al. 2015
M. filamentus	*Ictiobusbubalis*	Gills	13.1	16.3	7.8	6.2	
M. improvisus	*Leuciscusidus*	Muscles	6.5–7.7	7.5–9.3	2–3.3	-	
M. petenensis	*Dorosoma petenense*	circumorbital integument	11.8 (10.1–13.1)	13.8 (12–15.8)	7.1 (6.0–8.0)	5.3 (4.8–5.7)	Eiras et al. 2005
M. leafa	*Labeobata*	Gills	11.8–15.3 (19.1 ± 1.1)	13.6–15.3 (14.5 ± 0.6)	5.9–8.5 (6.7 ± 0.7)	5.1–5.9 (5.3 ± 0.3)	
M. omari	*Pangasianodon hypophthalmus*	Muscles	7.2–8.8 (7.9)	11.0–13.9 (12)	4.4–6.6 (5.9); 4.0–6.2 (5.6)	4.0–5.3 (4.7); 3.6–4.9 (4.3)	Eiras et al. 2014

Besides the comparison with host related species, the current *Myxobolus* species was matched with other morphologically similar species from different hosts. The present species shows similarities *M. brachysporus*; *M. filamentus*; *M. microlatus*; *M. improvisus*; *M. petenensis*; *M. leafa* and *M. omari*. It was revealed that *M. Brachysporus* (Baker 1963; Abdel-Bakiet al. 2015) from the spleen of different species of tilapiaare bigger in spore size and has smaller polar capsule than the current species. While in case of *M. filamentus*, spore size is bigger and has larger polar capsule in comparison to the present species. As well as the present species has slight difference in the polar capsules as bigger and smaller. In the present species there are two nearly equal polar capsules but *M. microlatus, M. improvises* and *M. petenensis* possess unequal polar capsule although *M. improvisus*is smaller in size whereas *M. microlatus* and *M. petenensis* capsules are bigger than the present species. In *M. leafa* polar capsules are oval, equal with prominent pointed tips at the posterior end just like a leaf (Thounaojam et al., 2013). The present species lack iodinophilous vacuole while *M. omari* has bigger spore size, an intercapsular process and iodinophilous vacuole (Székely et al 2009). In view of above differences from all closely related species, the present species is confirmed as a new species *M. deformis*sp. nov.

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